

CLAIMS

- 1- A DNA molecular size marker that contains DNA fragments which are 441, 325, 231, 210, 131, 116, 94 and 79 base pairs long (Figure 1-Marker B).
- 2- The production of molecular size marker in claim 1 is done by isolation of DNA from mycobacteriae, amplification of hsp65 gene by PCR, purification of DNA amplification products, molecular cloning, plasmid isolation and restriction enzyme digestion.
- 3- The species of mycobacteriae used for DNA isolation referred in claim 2 for production of the molecular size marker in claim 1, are the ones which produce the required size fragments indicated in claim 1.
- 4- The species of mycobacteriae referred in claim 3 which are used for production of molecular weight marker referred in claim 1 are *M. simiae*, *M. smegmatis*, *M. gallinarum*, *M. intracellulare*, *M. terrae*.
- 5- The primers used in amplification of hsp65 gene referred in claim 2 are TB11 (5' ACC AAC GAT GGT GTG TCC AT 3'), and TB12 (5' CTT GTC GAA CCG CAT ACC CT 3').
- 6- The restriction enzyme indicated in claim 2 is BstEII.
- 7- Molecular size marker indicated in claim 1 is used in determining the size of restriction fragments obtained by BstEII digestion, in the step of electrophoretic analysis of hsp65 PCR-REA (Polymerase Chain Reaction – Restriction Enzyme Analysis) method.
- 8- A DNA molecular size marker that contains DNA fragments which are 185, 161, 152, 139, 127, 103, 87, 69, 59, 58, 42, 40, 36 and 34 base pairs long (Figure 2- Marker H).
- 9- The production of molecular size marker in claim 8 is done by isolation of DNA from mycobacteriae, amplification of hsp65 gene by PCR, purification of DNA amplification products, molecular cloning, plasmid isolation and restriction enzyme digestion.
- 10- The species of mycobacteriae used for DNA isolation referred in claim 9 for production of the molecular size marker in claim 8, are the ones which produce the required size fragments indicated in claim 8.
- 11- The species of mycobacteriae referred in claim 10 which are used for production of molecular weight marker referred in claim 8 are *M. tuberculosis*, *M. simiae*, *M. gallinarum*, *M. chitae*, *M. xenopi*.
- 12- The primers used in amplification of hsp65 gene referred in claim 9 are TB11 (5' ACC AAC GAT GGT GTG TCC AT 3'), and TB12 (5' CTT GTC GAA CCG CAT ACC CT 3').
- 13- The restriction enzyme indicated in claim 9 is HaeIII.

14- Molecular size marker indicated in claim 8 is used in determining the size of restriction fragments obtained by HaeIII digestion, in the step of electrophoretic analysis of hsp65 PCR-REA (Polymerase Chain Reaction – Restriction Enzyme Analysis) method.